Trichothecenes: Natural Occurrence and Potential Hazard

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ABSTRACT

The trichothecene toxins are a chemical group of fungal metabolites characterized by a tetracyclic 12, 13-epoxy-trichothec-9-ene skeleton. There are over 40 naturally occurring derivatives produced predominantly by species of Fusarium but also produced by species of Cephalosporium, Myrothecium, Trichoderma, and Stachybotrys. The trichothecene derivatives most commonly encountered in feedstuffs are: T-2 toxin, diacetoxyscirpenol (DAS), deoxynivalenol (vomitoxin) and nivalenol. Deoxynivalenol has been isolated from corn and mixed feed more frequently than the other derivatives reported, and moreover, it is frequently found together with the fungal estrogen zearealenone. Deoxynivalenol has frequently been associated with corn refused by swine, vomiting in dogs, and corn used by humans in South Africa. Deoxynivalenol is produced by Fusarium roseum, an organism that is commonly found infecting corn in the field and corn stored on-the-cob in cribs in the Midwest. The trichothecenes have been implicated in diseases of man called alimentary toxic aleukia in the USSR and have been found in corn in the Transkei where the incidence of esophageal cancer is high. Among domestic animals, trichothecenes such as T-2 toxin, DAS and deoxynivalenol have been implicated in diseases characterized by a hemorrhagic syndrome in cows and swine, vomiting in swine and dogs, decrease in avian egg production and body weight, refusal of feed by swine and infertility in swine. The data on the residue of the administered T-2 toxin in domestic animals suggests that trichothecenes can be transmitted into humans.

INTRODUCTION

Fungi imperfecti of the genera Fusarium, Myrothecium, Trichoderma, Cephalosporium, Trichothecium, Verticillium, and Stachybotrys produce a group of chemically related toxic compounds called trichothecenes, characterized by a tetracyclic structure with a specific stereochemistry as shown in Figure 1. About 45 derivatives have been isolated from cultures of these fungi. Among those isolated, over 20 are known to be produced by species of Fusarium alone. Recently, baccharin, a very potent antileukemic trichothecene, has been isolated from a higher plant, Baccharis megapotamica (Asteraceae) (1).

The chemistry, biochemistry, and toxicology of trichothecenes have been extensively reviewed (2-6). The role of trichothecenes as an etiologic agent in mycotoxoses was first elucidated by a group of investigators at the University of Wisconsin (7). They demonstrated the presence of T-2 toxin, a trichothecene produced by F. tricinctum, in moldy corn associated with illness and death of lactating cows in a herd in Wisconsin. However, the involvement of trichothecenes in several sporadic outbreaks of mycotoxoses described as moldy corn toxicosis (7,8), alimentary toxic aleukia (ATA) (9), stachybotrytis toxicosis (10,11) and fescue foot disease (12) was suspected for some time (2) because the signs of those diseases resembled toxicity as incited by trichothecenes. Wyatt et al. (13,14) reported signs in poultry which were identical to those produced by T-2 toxin in laboratory tests and strongly suggested the actual presence of at least some trichothecene derivative. In another report, Eppley et al. (15) tested 173 samples of corn collected from various grain elevators in the Midwest and found that 94 (54%) contained a "skin irritating factor", presumably T-2 toxin, ranging in concentration from 0.05 to 1.0 µg/g of corn (0.05-1.0 ppm). The presence of T-2 toxin was not demonstrated, however.

There are a number of cases reported describing the occurrence of trichothecenes in feedstuffs naturally contaminated. In this paper those cases have been reviewed in an attempt to elaborate the role of trichothecenes in mycotoxicoses and to evaluate the potential hazards of these natural toxic contaminants.

Natural Occurrence

Evidence for involvement of any trichothecene in moldy corn toxicosis was considered to be circumstantial until Hsu et al. (7) found 2 ppm of T-2 toxin in moldy corn associated with a lethal toxicosis in dairy cattle. In Japan several outbreaks of "red mold toxicoses" of humans and animals were described (16) and attributed to ingestion of grain infected by Fusarium. Although no trichothecene was isolated from the grain actually involved in the intoxication, a sample of barley naturally infected with F. roseum was shown to be contaminated with two trichothecenes, deoxynivalenol and nivalenol in the concentrations of 6.23 and 7.15 ppm, respectively (17). During recent years in the midwestern United States, corn infected with F. roseum, as well as other species of Fusarium, caused multiple problems when fed to swine: (a) hyperestrogenism, (b) emesis, and (c) refusal of feed. Such corn was associated with wet weather that preceded harvest. Vesdonder et al. (18,19) identified deoxynivalenol as one of the trichothecenes present in the corn causing emesis and refusal of feed. Hibbs et al. (20) reported the presence of T-2 toxin in a commercial pellet feed involved in a case of hemorrhagic syndrome in cattle. A moldy grain toxicosis involving ducks, geese, horses and swine has been noted by Greenway and Pals (21) in British Columbia, Canada. The clinical signs described by them included emesis and refusal of the feed in swine, fever and excessive salivation in horses, and emesis, thirst and death in ducks and geese; visible lesions of the esophagus, proventriculus, and gizzards of geese were
also noted. This toxicity was linked to T-2 toxin present at a level of ca. 2.5 ppm in barley used in the feedstuffs (22), although some question was raised as to the authenticity of the toxin identified.

A number of undiagnosed cases of animal toxicoses pass through veterinary diagnostic laboratories, which have no explanation as to causality except perhaps that the feed might be implicated; the signs of intoxication include abortion, diarrhea, emesis, loss of weight gain, hemorrhagia and death. Routinely, these feed samples are analyzed for toxic fungi and toxic components. During the past four years, we analyzed over 200 such samples of feeds and feedstuffs associated with such toxicoses in animals. Trichothecene mycotoxins were identified in some of these samples and are presented in Table I (23).

Sample No. FS-382 and FS-404 were mixed feed obtained from the College of Veterinary Medicine of the University of Minnesota and were associated with an idiopathic condition in swine called hemorrhagic bowel syndrome (24). Each of the samples represented a separated incidence of this disease which occurred among experimental animals at the University. Diacetoxyscirpenol, another trichothecene produced by species of Fusarium, was isolated from these samples and was the only toxic component found. Weaver et al. (25) reproduced hemorrhagic bowel lesions in swine by the administration of pure diacetoxyscirpenol intravenously.

Sample No. 417 was associated with bloody stools in cattle; analyses revealed 78 ppb of T-2 toxin.

Four whole kernel corn samples (FS-356, 363, 398) and one commercial mixed feed sample (FS-463), which were refused by swine, were referred to our laboratory for analysis. The corn samples originated in Michigan, Indiana, and Ohio and the mixed feed in Minnesota. Analysis showed the presence of deoxynivalenol (vomitoxin) at levels from 50 to 1800 ppb (the percentage recovery was not estimated). The identity of deoxynivalenol was authenticated by mass spectroscopy. Although Vesonder et al. (18,19) reported deoxynivalenol in corn, this was the first report of its occurrence in a commercial feed sample. All samples analyzed also contained zearalenone; isolates of F. roseum which produce deoxynivalenol usually synthesize zearalenone.

Samples consisting of hand-selected 100% moldy maize kernels (FS-516B) from 1973/74 Zambian crop and from maize harvested in South Africa (FS-516A) were found to be heavily contaminated with deoxynivalenol and zearalenone (26).

A corn sample (FS-543B) associated with intoxication in cows was submitted by the Veterinary Clinic in Arizona. The clinical signs described were central nervous system irritations, muscle tremors, irregular heat cycles and even death. We found the sample to be contaminated with deoxynivalenol; zearalenone was also detected in this sample. Samples FS-570A-D associated with feed refusal and emesis were contaminated with deoxynivalenol and zearalenone; the sample FS-570A which had been totally refused by swine was found to contain rather low amounts of deoxynivalenol.

Deoxynivalenol and zearalenone were also found in maize samples in some areas in the Transkei. These samples were collected in the areas of incidences of esophageal cancer in human, and were naturally contaminated with Fusarium (W.F.O. Marasas, private communication).

Recently, Siegfried (27) reported that he isolated 31.5 ppm of diacetoxyscirpenol from maize (used as feed); however, a detailed account of this occurrence is not given.

### Potential Hazards

Toxicological problems in humans and animals associated with consumption of moldy grains have long been recognized; and those particularly suspected of being trichothecene-toxics are listed in Table II. Moldy corn toxicosis as described by Hsu et al. (7) and Smalley et al. (8) in northern climates is associated with many different fungi, predominant among which is F. tricinctum. The signs of this disease are similar to those described by Forcag (10), namely general digestive disorders, bloody diarrhea, and hemorrhagic lesions in the stomach, heart, intestine, lungs, bladder and kidneys.

ATA in man was described by Joffe in the Soviet Union (9). This disease was associated with moldy millet primarily, but also with wheat, rye, oats, and buckwheat. The signs described were "typical spots on the skin, leukopenia, agranulocytosis, necrotic angina, hemorrhagic diathesis, sepsis and exhaustion of the bone marrow." Of the fungi involved, F. sporotrichioides (syn. F. tricinctum) and F. poae were thought to be involved, and the toxins were described as psor fusarin and spor fusarin, steroidal compounds. More than 10% of the population in the Orenburg district of the Soviet Union was affected at one time.

The symptoms and signs of this disease resembled toxicity of the trichothecenes as reported in the United States. An authentic sample of psor fusarin was obtained and analyzed for the steroids. Modern analytical methods were used to identify the constituents, but no trace of a steroid could be found. However, 2.5% of the sample was...